Food Chemistry 110 (2008) 1036–1040

Contents lists available at ScienceDirect

Food Chemistry

journal homepage: www.elsevier.com/locate/foodchem

Analytical Methods

Effect of gamma irradiation on the stability of anthocyanins and shelf-life of various pomegranate juices

H. Alighourchi, M. Barzegar *, S. Abbasi

Department of Food Science and Technology, Tarbiat Modares University, P.O. Box 14115-336, Tehran, Iran

ARTICLE INFO

Article history:
Received 29 September 2007
Received in revised form 18 February 2008
Accepted 4 March 2008

Keywords:
Food irradiation
Ionizing radiations
Pomegranate juice
Anthocyanin
Fungi

ABSTRACT

Food irradiation is a process which exposing food to ionizing radiations and it can improve the safety of food. The pomegranate juice contained considerable anthocyanins and has become a new functional food available for dieting and health. In the present study, the effects of gamma irradiation (0–10 kGy) on the stability of anthocyanins and inhibition of microbial growth in pomegranate juice during storage were investigated. Results indicated that the irradiation at all applied doses, significantly reduced total and individual anthocyanins. Moreover, irradiation with higher dosages (3.5–10 kGy) had undesirable effect on the total content of anthocyanins. However, irradiation at 2.0 kGy had effectively diminished the total bacteria and fungi count and retarding microbial growth during storage. Based on adverse effect of gamma irradiation on ACs content of studied juices, it is not recommended to irradiate pomegranate juice with dosage higher than 2.0 kGy.

© 2008 Elsevier Ltd. All rights reserved.

1. Introduction

Pomegranate (*Punica granatum* L.) is one of the oldest known edible fruits and is grown in Afghanistan, China, India, Iran, Japan, Mediterranean countries, Russia and the US (Shahidi & Naczk, 2004). In Iran, pomegranate is eaten as fresh as well as processed for jams, jellies, syrups and pomegranate juice products. This fruit is one of the most important commercial fruits in Iran and its total production in year 2005 was ~670,000 tons (Anonymous, 2005).

Anthocyanins (ACs) are glycosides of polyhydroxy and polymethoxy derivatives of flavylium cation. Recent studies demonstrated that polyphenolic flavonoids exhibit a wide range of biological, pharmacological and chemoprotective properties as free radical scavengers preventing oxidation and cancer initiation (Kong, Chia, Goh, Chia, & Brouillard, 2003).

Based on some research, pomegranate juice is a rich source of different phenolic compounds, with ACs to be one of the most important classes (Du, Wang, & Francis, 1975). The main ACs of pomegranate arils identified were as delphinidin 3,5-*O*-diglucoside (Dp 3,5-*O*-dG), 3-*O*-glucoside (Dp 3-*O*-G), cyanidin 3,5-*O*-diglucoside (Cy 3,5-*O*-dG), 3-*O*-glycoside (Cy 3-*O*-G), pelargonidin 3,5-*O*-diglycoside (Pg 3, 5-*O*-dG), and 3-*O*-glycoside (Pg 3-*O*-G) (Martí, Pérez-Vicente, & García-Viguera, 2000; Miguel, Dundlen, Antunes, Neves, & Martins, 2004). Pomegranate ACs are labile compounds and easily susceptible to degradation during storage and processing. Martí et al. (2000) found that the total ACs content was reduced to 20% after storage of pomegranate juice at 5 °C for 6 months. In addition, Miguel et al. (2004) observed that ACs content of *Assaria* variety of pomegranate decrease significantly during 72 h storage at 4 °C.

In the recent years, the tendency of consumers to fresh fruit juices has been increased due to their better organoleptic properties than pasteurized ones. Previously, it was generally assumed that pathogenic microorganisms could not survive in high acid foods. However, recent outbreaks of foodborne illnesses from unpasteurized fruit juices have indicated the necessity of pasteurization for all fruit juices (Parish, 1997). Microorganisms, in particular acid tolerant bacteria and fungi such as yeasts and molds, can easily spoil fruit juices (Tournas, Heeres, & Burgess, 2006). During the last decade, with increasing demand for nutritious, fresh-like food products with high organoleptical quality and an adequate shelf-life, non-thermal inactivation techniques have been a major research issue. Some of the investigated technologies are irradiation, high hydrostation pressure (HHP), pulsed electrical fields, and UV decontamination (Devlieghere, Vermeiren, & Debevere, 2004). Preservation and shelf-life extension have been the historical focus of research on irradiation of juices (Niemira & Deschênes, 2004, chap. 11). Food irradiation is a means of food preservation that has been in development since the early part of the 20th century. If applied properly, irradiation can be an effective way to reduce the incidence of foodborne diseases and also inactivates food

* Corresponding author. Tel.: + 98 21 44196522; fax: +98 21 44196524.
E-mail addresses: mbb@modares.ac.ir, mohsenbb@gmail.com, barzegarm@yahoo.com (M. Barzegar).

0308-8146/$ - see front matter © 2008 Elsevier Ltd. All rights reserved.
doi:10.1016/j.foodchem.2008.03.013

spoilage organisms, including bacteria, molds, and yeasts in our food supply (Morehouse, 2002). The FAO/IAEA/WHO joint committee on the wholesomeness of irradiated food approved irradiation technology in 1981. It was stated that, irradiation of food at doses up to 10 kGy introduced no special nutritional problem (Stevenson, 1994).

The key organisms in food irradiation were yeasts and molds, which have a higher D_{10} value than bacterial pathogens (Monk, Beuchat, & Doyle, 1994). In fruit juices, D_{10} values have been reported for yeasts and molds are in the range of 1–3 kGy (Narvaiz, Lescano, & Kairiyama, 1992) and 0.3–0.7 kGy for pathogenic bacteria (Buchanan, Edelson, Snipes, & Boyd, 1998; Niemira, Sommers, & Boyd, 2001). Buchanan et al. (1998) showed that, a dose of 1.8 kGy should be sufficient to achieve the 5D inactivation of *Escherichia coli* recommended by the National Advisory Committee for Microbiological Criteria for Foods irradiation. In addition, Chachin and Ogata (1969) investigated the effect of sterilizing (2–80 kGy) doses of gamma irradiation in grape juice.

Based on our knowledge, there is no information about the anthocyanin contents of major varieties of Iranian pomegranates and their variations after gamma irradiation. Therefore, the objectives of the present study were as follows: (i) identifying main ACs of some pomegranate varieties; (ii) subjecting pomegranate juices to various levels of gamma irradiating and investigating its effect on ACs content and microbiological properties or shelf-life; and (iii) investigating the effect of pomegranate variety on ACs content of juices after gamma irradiation.

2. Materials and methods

2.1. Samples

Six various pomegranate varieties were obtained from mature fruits growing in agricultural research center of Yazd in south west of Iran. Commercially ripe fresh fruits were harvested during the September of 2006 from different mature trees and they were randomly selected to represent the population of the plantation. Fruits were transported by ventilated car to the laboratory, where pomegranates with defects (sunburns, cracks, cuts and bruises in husk) were discarded. The different varieties selected for this study were as follows: *Gorche Shahvar Yazdi* (GSY), *Malase Yazdi* (MY), *Vahshe Kane Tehran* (VKT), *Mesri Torshe Kazeron* (MTK), *Jangali Pust Germeze Rodbare Torsh* (JPGRT) and *Torshe Mamoli Lasjer* (TML). Approximately, 2 kg (n = 10) of pomegranates at harvesting maturity was sampled for each variety.

2.2. Chemicals

Delphinidin 3,5-*O*-diglucoside (Dp 3,5-*O*-dG), delphinidin 3-*O*-glucoside (Dp 3-*O*-G), cyanidin 3,5-*O*-diglucoside (Cy 3,5-*O*-dG), cyanidin 3-*O*-glucoside (Cy 3-*O*-G), pelargonidin 3,5-*O*-diglucoside (Pg 3,5-*O*-dG), and pelargonidin 3-*O*-glucoside (Pg 3-*O*-G) standards were purchased from Apin Chemicals Co. Ltd. (Oxfordshire, UK). Methanol (HPLC grade) was purchased from Caledon laboratories (Ontario, Canada). Formic acid and all bacteriological media were obtained from Merck (Darmstadt, Germany). The ultra pure water was prepared with the Purise system (Seoul, South Korea).

2.3. Preparation of raw pomegranate juice

Fruit from each pomegranate variety was washed in cold tap water and drained. They were manually cut-up and the outer leathery skin, which encloses hundreds of fleshy sacs, was removed. The arils were manually separated and pressed. The juices (50 mL) were centrifuged (2 min at 10,000 rpm at −4 °C), and divided into small vials and kept frozen at −18 °C for one day upon analysis.

2.4. Irradiation treatment

The pomegranate juices were filled in sterilized amber Wheaton-320 glass vials (8 mL, from Sigma, USA). Irradiation treatment was carried out at various doses (0, 0.5, 2.0, 3.5, 5.0 and 10.0 kilogray (kGy)) at a dose rate of 1.43 kGy/h using a Gamma cell-220 irradiator (Nordion, Canada). After gamma irradiation, the samples were immediately placed in refrigerator at 4 °C and they were immediately analyzed by HPLC/UV. Three samples were prepared for each treatment.

2.5. Microbiological analysis

The enumeration of microorganisms was made using standard techniques (AOAC, 1984), and included plate counts, yeasts and molds and coliform. Microbial analyses of treated and untreated pomegranate juices were determined during two weeks storage at 4 °C (1, 2, 3, 7, and 14 days). Serial dilutions of original samples were prepared in sterile peptone water (0.1%) to reduce the microbial population sufficiently to obtain separate colonies when plating. Total plate counts were determined using pour plate method on Plate Count Agar. One milliliter of several diluted samples were mixed with 15 mL liquid plate count agar that had been cooled to about 45 °C, and poured immediately into sterile 90 mm Petri dishes. After the agar had hardened, incubation was performed at 35 °C for 48–72 h. Total fungi (moulds and yeasts) were carried out in triplicate using pour plate method on plates of Potato Dextrose Agar (PDA) medium, and then were enumerated after incubation for 5 days at 25 °C. For a coliform count, samples were plated directly onto MacConkey agar and incubated at 37 °C for 24–36 h. Each value represents the mean of three samples and results were expressed as colony-forming units (CFU) per milliliter.

2.6. Anthocyanin analysis

ACs content of juices were determined using a Waters HPLC system equipped with an Empower software, a pump (Waters 600, USA), a Rheodyne 7125i six-way injector with 20 μL sample loop, and a UV–Vis detector (Waters model 2487). A column μBondapack^{TM} C_{18} (4.6 × 250 mm, dp 10 μm) from Waters (Ireland)) was used for the separation.

Clarified juice (20 μL) was injected onto the HPLC. The elution was carried out at room temperature using 5% formic acid aqueous solution (A) and methanol (B) in a linear gradient from 15% to 35% B at 15 min, followed by isocratic run until 20 min. Flow rate was 1 mL/min with UV–Vis detector at 510 nm (Miguel et al., 2004). Calculation of the concentrations was based on the external standard method and ACs were identified by comparison of their retention times with those of pure standards (Table 1). For each sampling point, there were three replicates.

2.7. Statistical analysis

One-way analysis of variance was used to analyze the data. A p value of 0.05 or less was considered as significant (using SAS software).

Table 1
Linear calibration equations of individual anthocyanin standards

Compound	t_R (min)	Linear range (mg/L)	Linear equation	r
Dp 3, 5-*O*-dG	9.9	1–100	$A = 18718C - 41455$[a]	0.9995
Cy 3, 5-*O*-dG	10.6	1–500	$A = 18720C - 57351$	0.9998
Pg 3, 5-*O*-dG	11.2	0.5–50	$A = 23444C + 431$	1.00
Dp 3-*O*-G	11.9	1–250	$A = 22933C - 47821$	0.9999
Cy 3-*O*-G	12.7	1–250	$A = 76950C - 439615$	0.9989
Pg 3-*O*-G	13.5	0.02–50	$A = 66468C - 17320$	0.9995

[a] A: peak area; C: concentration (mg/L).

3. Results and discussion

3.1. Effect of irradiation on ACs content

Based on our findings, some of 6 different ACs, including delphinidin 3,5-*O*-diglucoside, 3-*O*-glucoside, cyanidin 3,5-*O*-diglucoside, 3-*O*-glucoside, pelargonidin 3,5-*O*-diglucoside, and 3-*O*-glucoside were identified in freshly prepared juices by comparing their retention times with those of authentic standards (and by spiking), which confirm previously reports on the other pomegranate varieties (Martí et al., 2000; Miguel et al., 2004).

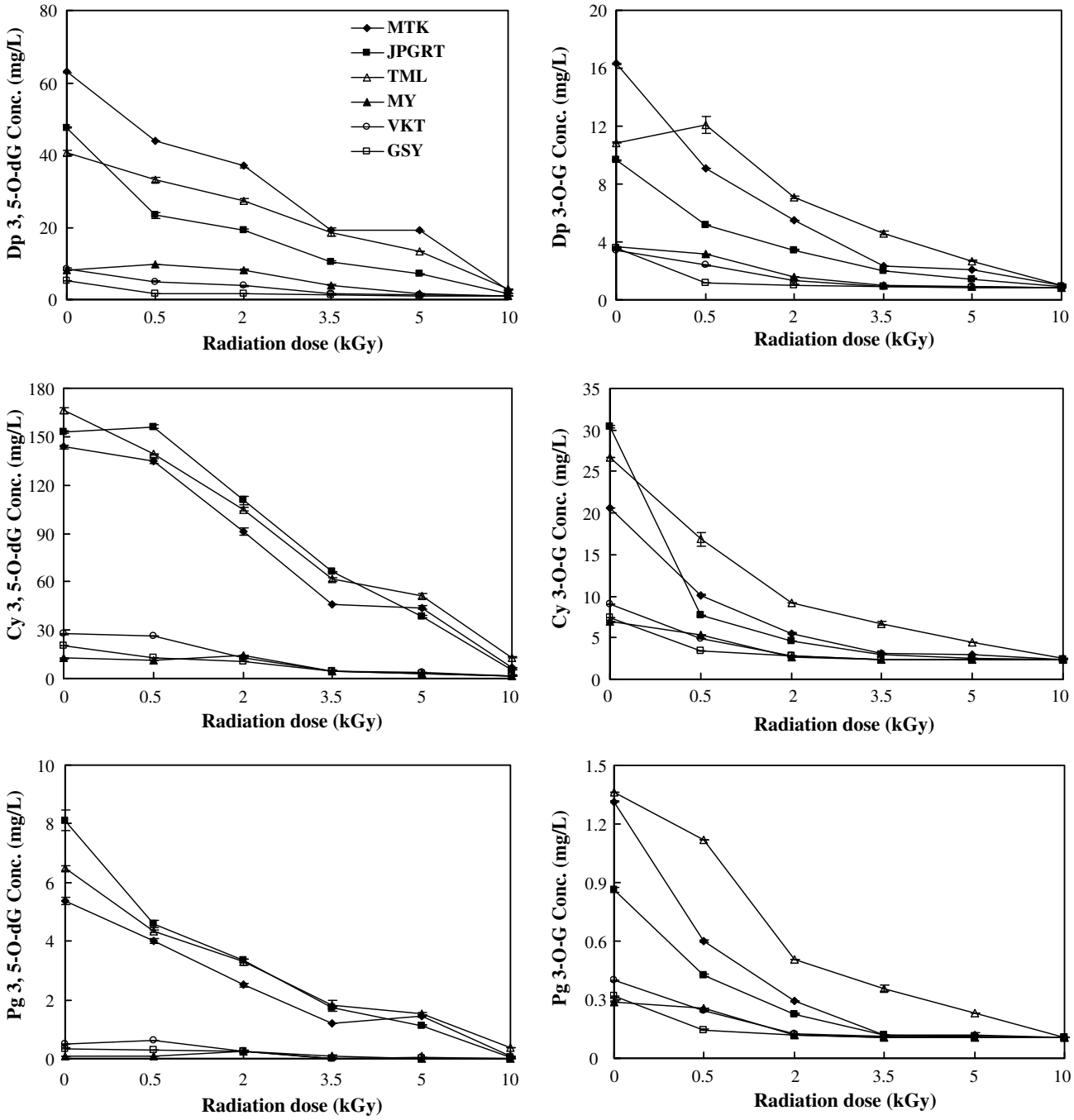

Fig. 1. Evolution of the individual anthocyanin concentrations of control (0 kGy) and irradiated (0.5–10 kGy) pomegranate juices.

Irradiation was carried out at several doses to find the optimal dose for getting maximum microbial reduction and minimum degradation of ACs as the color of pomegranate juice is directly depended on its content and composition. The current study appears, is one of the only ones that examined the effect of irradiation by different doses on the AC contents and microbiological behavior of pomegranate juices. Although there are many studies on the effect of irradiation on microbiological and organoleptic attributes of other fruit and vegetable juices (Buchanan et al., 1998; Chachin & Ogata, 1969; Kim, Song, Lim, Yun, & Chung, 2007; Song et al., 2006).

Fig. 1 shows that gamma irradiation of studied juices cause a significant decrease on the individual ACs concentration in comparison with the initial AC contents (dose 0 kGy) due to degradation of ACs ($p < 0.05$). As it can be seen, the stability of diglycosides ACs to irradiation were higher than monoglycosids at lower doses of gamma irradiation (0.5 and 2 kGy). For example, cyanidin 3,5-*O*-diglucoside was more stable (up to 2 kGy) than the other ACs. However, decreasing trends of diglycosides were similar to monoglycosides in the higher level of gamma irradiation. In addition, higher stability of diglycosides ACs at different conditions were previously reported by other researchers (Holcroft, Gil, & Kader, 1998; Timberlake & Bridle, 1977).

Table 2 shows total AC contents of all studied pomegranates before and after irradiation by gamma ray (0–10 kGy). It can be seen that the different applied doses had significant influence on the total ACs contents of all juices after irradiation ($p < 0.05$). Chachin and Ogata (1969) have previously investigated the effect of sterilizing (2–80 kGy) doses of gamma irradiation in grape, apple, and orange juices. They reported that gamma irradiation, at $\geqslant 10 \leqslant 80$ kGy, obviously caused losing of grape juice ACs. In the irradiated kale juice (0, 3, and 5 kGy), the amount of total phenolic content was significantly lower than the control immediately after the irradiation (Song et al., 2006). In contrast, Ayed, Yu, and Lacroix (1999) investigated the effect of irradiation (0–9 kGy) of grape pomace and they observed that AC contents increased at all doses of irradiation, especially at 6 kGy.

In our study, irradiation at 5 and 10 kGy did not show significant difference ($p \geqslant 0.01$) between the amounts of total ACs of MY, VKT and GSY varieties. The average loss of the total AC contents in all juices, after gamma irradiation (0–10 kGy), were between 22.0% and 90.0%. In fact, pomegranate juices that irradiated at 0.5 and 10 kGy retained approximately 80% and 10% of the AC content compared with the control, respectively. These results indicated that the stability of ACs against the irradiation was related to juice composition. The differences among juices originated from fruits variety, and the relative stability of an AC depends on its matrix, structural features, and the processing conditions (Es-Safi, Cheynier, & Moutounet, 2002; Talcott, Brenes, Pires, & Del Pozo-Insfran, 2003; Torskangerpoll & Andersen, 2005). However, there are few pieces of research in the literature about the effect of the fruit variety on the radiation sensitivity of the resulting juices.

3.2. Effect of irradiation on microbial flora

The predominant organisms found in the various types of juices were yeasts (Hatcher, Parish, Weih, Splittstoesser, & Woodward, 2000; Tournas et al., 2006). These organisms could grow during refrigeration (4 °C) and cause spoilage of samples. In food irradiation, the D_{10} values of yeasts and molds were higher than bacterial pathogens. Thus, most research efforts related to irradiation of juices have targeted spoilage organisms such as yeasts and molds rather than bacterial pathogens (Grinbaum, Ashkenazi, Treister, Goldschmied-Requven, & Block, 1994; Monk et al., 1994).

Table 2
Total anthocyanin contents of irradiated and non-irradiated pomegranate juices (mg/L)

Irradiation dose (kGy)	Pomegranate varieties[a]					
	MTK	JPGRT	TML	MY	VKT	GSY
0	251.7 ± 0.1au	249.2 ± 0.6bu	252.2 ± 1.0au	31.7 ± 0.6eu	49.4 ± 0.1cu	37.2 ± 0.0du
0.5	202.7 ± 1.8bv	197.3 ± 0.0cv	206.7 ± 0.3av	30.2 ± 0.5ev	39.4 ± 0.4dv	19.4 ± 0.0fv
2	142.1 ± 2.8bw	141.2 ± 2.3bw	152.6 ± 0.4aw	27.2 ± 0.3cw	21.4 ± 0.3dw	16.3 ± 0.1ew
3.5	72.2 ± 0.3cx	78.2 ± 0.4bx	98.0 ± 3.0ax	12.6 ± 0.5dx	9.5 ± 0.0ex	9.0 ± 0.1ex
5	69.5 ± 1.0bx	50.5 ± 0.5cy	73.1 ± 1.8ay	8.4 ± 0.1dy	8.6 ± 0.1dy	7.0 ± 0.1dy
10	12.9 ± 0.2by	10.7 ± 0.1cz	20.0 ± 0.4az	6.0 ± 0.0dz	6.2 ± 0.1dz	6.1 ± 0.2dz

[a] Values with different letters (a–d) within a similar row and (w–z) within a similar column are significantly different ($p < 0.05$).

Table 3
Total plate counts (TPC) and total fungal counts (TFC) of irradiated and non-irradiated MY and TML pomegranate juices, stored at 4 °C for 14 days[a]

Juice	Microbial counts (cfu/mL)	Irradiation dose(kGy)	Storage (days)				
			1	2	3	7	14
MY	TPC	0	4700 ± 145	7470 ± 850	19,850 ± 450	89,800 ± 800	536,000 ± 9200
		0.5	840 ± 30	1250 ± 140	2400 ± 380	19,800 ± 1050	49,250 ± 2500
		2	0	0	<10	27 ± 2	240 ± 25
	TFC	0	3400 ± 80	6900 ± 450	10,150 ± 1150	185,500 ± 7200	715,000 ± 3540
		0.5	1050 ± 230	2820 ± 190	4240 ± 380	25,180 ± 1350	87,200 ± 8540
		2	0	<10	0	14 ± 5	43 ± 12
TML	TPC	0	2060 ± 450	4200 ± 430	8250 ± 690	38,600 ± 4800	303,800 ± 7800
		0.5	398 ± 45	675 ± 90	2850 ± 150	9650 ± 1720	30,640 ± 2650
		2	0	0	20 ± 5	0	70±17
	TFC	0	1020 ± 440	3460 ± 560	7820 ± 1840	45,800 ± 5800	39,6450 ± 5250
		0.5	590 ± 80	1080 ± 45	2740 ± 240	11,850 ± 1500	42,890 ± 3300
		2	0	0	0	38 ± 12	85±32

[a] At >3.5 kGy, there were no microorganisms in MY and TML juices.

The changes of microbial populations of MY and TML (pH 4.2 and 4.1, respectively) pomegranate juices are shown in Table 3. The microbial counts (bacteria and fungi) were measured by the plant counts and total fungi in control and irradiated pomegranate juices. The initial mean populations of the total bacterial and fungi counts for fresh juices of MY and TML (without treatment) were $\sim 4.7 \times 10^3$, $\sim 3.4 \times 10^3$ and $\sim 2.0 \times 10^3$, $\sim 1.0 \times 10^3$ cfu/mL, respectively. After irradiation and during storage, no coliform was detected. The total counts of the bacteria in the non-irradiated juices of MY and TML, were rapidly increased to more than 5.3×10^5 and 3.0×10^5 cfu/mL, respectively, during storage at 4 °C. Gamma irradiation has prevented the microbial growth and by increasing the irradiation dosage a decreasing trend was observed (in studied varieties). Irradiation at 0.5 and 2 kGy reduced the growth rate of bacteria and fungi of the selected pomegranate juices during the first 3 days of storage at 4 °C. The microbial population reduced to below the detection limits at $\geqslant 3.5$ kGy, in all studied juices. Similar results were observed in other pomegranate varieties (GSY, VKT, MTK, JPGRT, data not shown). Based on the results of Aziz & Moussa, 2002 and Kim et al. (2007), the 3–5 kGy radiation doses may prevent microbial growth in the kale juice during storage period. This result showed that, the inactivation of microorganisms in different juices depended on their compositions. Buchanan et al. (1998) reported that differences among the juices may be affecting the radiation resistance of microorganism. Their results showed that low-dose gamma radiation treatment potentially could be eliminating *E. coli* without any detectable effect on the organoleptic quality of the product. In our study, we found that irradiation doses upper than 2 kGy were sufficient to completely inactivate the studied microorganisms.

4. Conclusions

Gamma irradiation can be applied to produce fruit juices with improved shelf-life. The different applied gamma doses significantly affected the total and individual ACs content of all juices and therefore pomegranate juice color. In fact, pomegranate juices that irradiated at 0.5 and 10 kGy retained approximately 80% and 10% of the initial AC contents. Irradiation at doses upper than 2 kGy can completely inactivate studied microorganisms as well as retarding microbial growth during storage. However, at higher doses (>2 kGy), a considerable decrease of total AC content was observed. Finally, it is not recommended to irradiate pomegranate juice with dosage higher than 2.0 kGy.

Acknowledgments

The authors would like to acknowledge Research Council of Tarbiat Modares University for its financial support and Agricultural Research Center of Yazd for providing fresh pomegranates.

References

Anonymous (2005). Iran Statistical Year Book 2005. <http://eamar.sci.org.ir/index_e.aspx>.

AOAC (1984). *Bacteriological Analytical Manual* (6th ed.). Washington, DC: Association of Official Analytical Chemists.

Ayed, N., Yu, H. L., & Lacroix, M. (1999). Improvement of anthocyanin yield and shelf-life extension of grape pomace by gamma irradiation. *Food Research International, 32*, 539–543.

Aziz, N. H., & Moussa, L. A. A. (2002). Influence of gamma-radiation on mycotoxin producing moulds and mycotoxins in fruits. *Food Control, 13*, 281–288.

Buchanan, R. L., Edelson, S. G., Snipes, K., & Boyd, G. (1998). Inactivation of *Escherichia coli* O157:H7 in apple juice by irradiation. *Applied and Environmental Microbiology, 64*(11), 4533–4535.

Chachin, K., & Ogata, K. (1969). Changes of chemical constituents and quality in some juice irradiated with the sterilizing dose level of gamma rays. *Food Irradiation, 4*(1), 85–90.

Devlieghere, F., Vermeiren, L., & Debevere, J. (2004). New preservation technologies: Possibilities and limitations. *International Dairy Journal, 14*, 273–285.

Du, C. T., Wang, P. L., & Francis, F. J. (1975). Anthocyanins of pomegranate, *Punica granatum*. *Journal of Food Science, 40*, 417–418.

Es-Safi, N., Cheynier, V. R., & Moutounet, M. (2002). Role of aldehydic derivatives in the condensation of phenolic compounds with emphasis on the sensorial properties of fruit-derived foods. *Journal of the Science of Food and Agriculture, 50*, 5571–5585.

Grinbaum, A., Ashkenazi, I., Treister, G., Goldschmied-Requven, A., & Block, C. S. (1994). Exploding bottles: eye injury due to yeast fermentation of an uncarbonated soft drink. *British Journal of Ophthalmology, 78*(11), 883.

Hatcher, W. S., Parish, M. E., Weih, J. L., Splittstoesser, D. F., & Woodward, B. B. (2000). Fruit beverages. In F. P. Downes & K. Ito (Eds.), *Methods for the Microbiological Examination of Foods* (pp. 565–568). Washington, DC: American Public Health Association.

Holcroft, D. M., Gil, M. I., & Kader, A. A. (1998). Effect of carbon dioxide on anthocyanins, phenylalanine ammonia lyase and glucosyltransferase in the arils of stored pomegranates. *Journal of the American Society for Horticultural Science, 123*, 136–140.

Kim, D., Song, H., Lim, S., Yun, H., & Chung, J. (2007). Effects of gamma irradiation on the radiation-resistant bacteria and polyphenol oxidase activity in fresh kale juice. *Radiation Physics and Chemistry, 76*, 1213–1217.

Kong, J. M., Chia, L. S., Goh, N. K., Chia, T. F., & Brouillard, R. (2003). Analysis and biological activities of anthocyanins. *Phytochemistry, 64*, 923–933.

Martí, N., Pérez-Vicente, A., & García-Viguera, C. (2000). Influence of storage temperature and ascorbic acid addition on pomegranate juice. *Journal of the Science of Food and Agriculture, 82*(2), 217–221.

Miguel, G., Dundlen, S., Antunes, D., Neves, A., & Martins, D. (2004). The effect of two methods of pomegranate juice extraction on quality during storage at 4 °C. *Journal of Biomedicine and Biotechnology, 5*, 332–337.

Monk, J. D., Beuchat, L. R., & Doyle, M. P. (1994). Irradiation inactivation of food-borne microorganisms. *Journal of Food Protection, 58*(2), 197–208.

Morehouse, K. M. (2002). Food irradiation – US regulatory considerations. *Radiation Physics and Chemistry, 63*, 281–284.

Narvaiz, P., Lescano, G., & Kairiyama, E. (1992). Irradiation of almonds and cashew nuts. *Lebensmittel-Wissenschaft und-Technologie, 25*, 232–235.

Niemira, B. A., & Deschênes, L. (2004). Ionizing radiation processing of fruits and fruit products. In D. M. Barrett, L. Somogyi, & H. Ramaswamy (Eds.), *Processing fruits* (2nd ed.). Boca Raton, FL: CRC press.

Niemira, B. A., Sommers, C. H., & Boyd, G. (2001). Irradiation inactivation of four Salmonella species in orange juices with varying turbidity. *Journal of Food Protection, 64*(5), 614–617.

Parish, M. E. (1997). Public health and nonpasteurized fruit juices. *Critical Reviews in Microbiology, 23*(2), 109–119.

Shahidi, F., & Naczk, M. (2004). Phenolic compounds in fruits and vegetables. In *Phenolic in food and nutraceuticals* (pp. 131–239). Boca Raton, FL: CRC Press.

Song, H. P., Kim, D. H., Jo, C., Lee, C. H., Kim, K. S., & Byun, M. W. (2006). Effect of gamma irradiation on the microbiological quality and antioxidant activity of fresh vegetable juice. *Food Microbiology, 23*, 372–378.

Stevenson, M. H. (1994). Nutritional and other implications of irradiating meat. *Proceedings of the Nutrition Society, 53*, 317–325.

Talcott, S. T., Brenes, C. H., Pires, D. M., & Del Pozo-Insfran, D. (2003). Phytochemical stability and color retention of copigmented and processed muscadine grape juice. *Journal of Agricultural and Food Chemistry, 51*, 957–963.

Timberlake, C. F., & Bridle, P. (1977). Anthocyanins: colour augmentation with atechin and acetaldehyde. *Journal of the Science of Food and Agriculture, 28*, 539–544.

Tournas, V. H., Heeres, J., & Burgess, L. (2006). Moulds and yeasts in fruit salads and fruit juices. *Food Microbiology, 23*, 684–688.

Torskangerpoll, K., & Andersen, Q. M. (2005). Colour stability of anthocyanins in aqueous solutions at various pH values. *Food Chemistry, 89*, 427–440.